Foxo3 Cas9-KO *Strategy*

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Project Overview

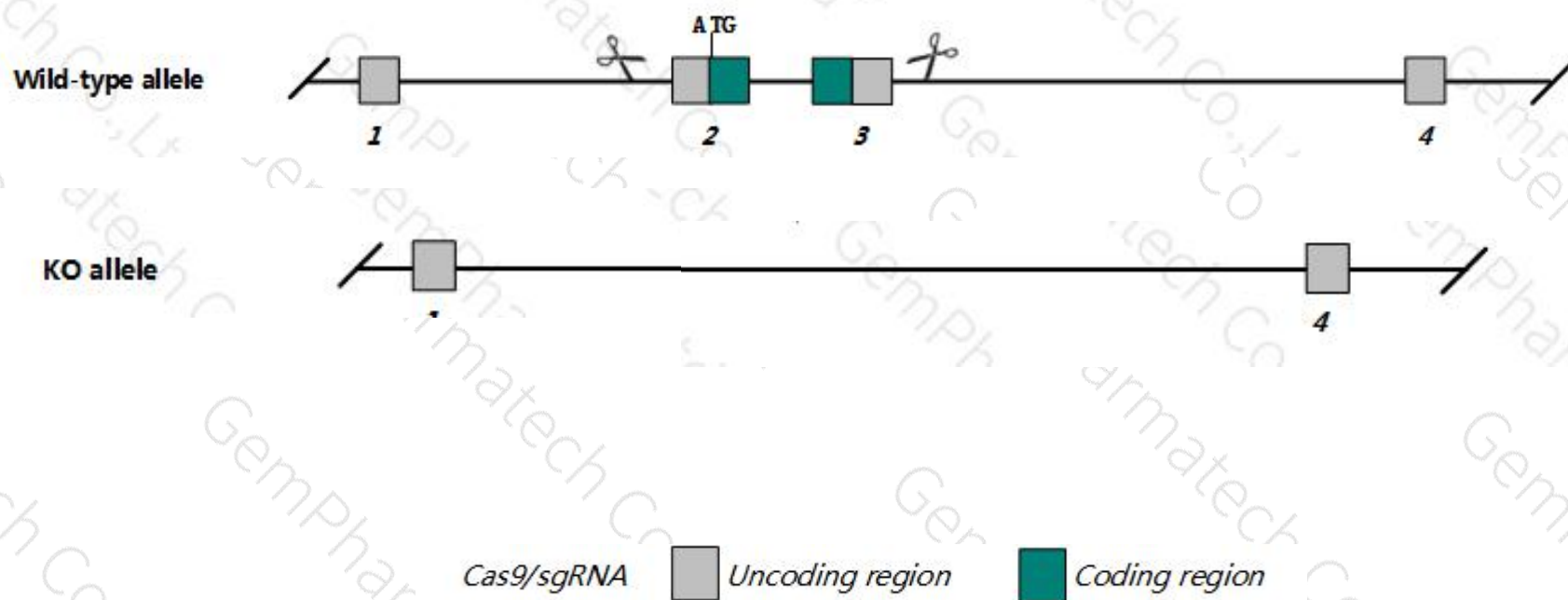|	
Project Name	*Foxo3*
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Foxo3* gene. The schematic diagram is as follows:

Technical routes

- The *Foxo3* gene has 5 transcripts, According to the structure of *Foxo3* gene, exon2-3 of *Foxo3*-201 transcript is recommended as the knockout region.The region contains all coding seqence.Knock out the region,result in destruction of protein.

- In this project we use CRISPR/Cas9 technology to modify *Foxo3* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating Positive F0 generation mice with C57BL/6JGpt mice.

Notice

➢ According to the existing MGI data: Inactivation of the locus results in an ovarian defect involving follicular growth activation and leads progressively to female sterility. For some alleles defects in immune system function and hematopoiesis have also been reported.

➢ The *Foxo3* gene is located on the Chr10. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.

➢ This Strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes, all risk of the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

Foxo3 forkhead box O3 [*Mus musculus* (house mouse)]

Gene ID: 56484, updated on 17-Oct-2018

Summary

|	
Official Symbol	Foxo3 provided by MGI
Official Full Name	forkhead box O3 provided by MGI
Primary source	MGI:MGI:1890081
See related	Ensembl:ENSMUSG00000048756 Vega:OTTMUSG00000020929
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Fkhr2; C76856; FKHRL1; Foxo3a; 1110048B16Rik; 2010203A17Rik
Expression	Ubiquitous expression in colon adult (RPKM 26.1), lung adult (RPKM 19.7) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

The gene has 5 transcripts, and all transcripts are shown below:

Name ▲	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags		
Foxo3-201	ENSMUST00000056974.3	2889	672aa	Protein coding	CCDS23810⟳	Q9WVH4⟳	TSL:1	GENCODE basic	APPRIS P1
Foxo3-202	ENSMUST00000105501.1	1316	244aa	Protein coding	-	H9KUZ0⟳	TSL:1	GENCODE basic	
Foxo3-203	ENSMUST00000105502.7	6848	672aa	Protein coding	CCDS23810⟳	Q9WVH4⟳	TSL:1	GENCODE basic	APPRIS P1
Foxo3-204	ENSMUST00000175881.7	2535	672aa	Protein coding	CCDS23810⟳	Q9WVH4⟳	TSL:1	GENCODE basic	APPRIS P1
Foxo3-205	ENSMUST00000177542.1	514	103aa	Protein coding	-	H3BKE2⟳	CDS 3' incomplete	TSL:3	

The strategy is based on the design of *Foxo3*-201 transcript,The transcription is shown below

< Foxo3-201
protein coding
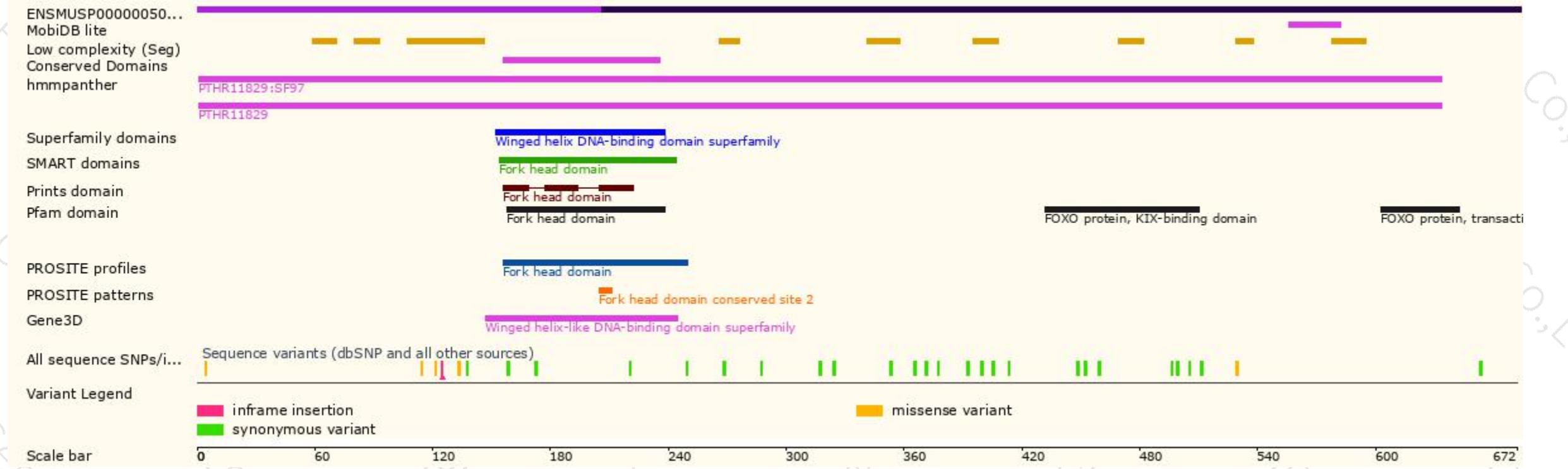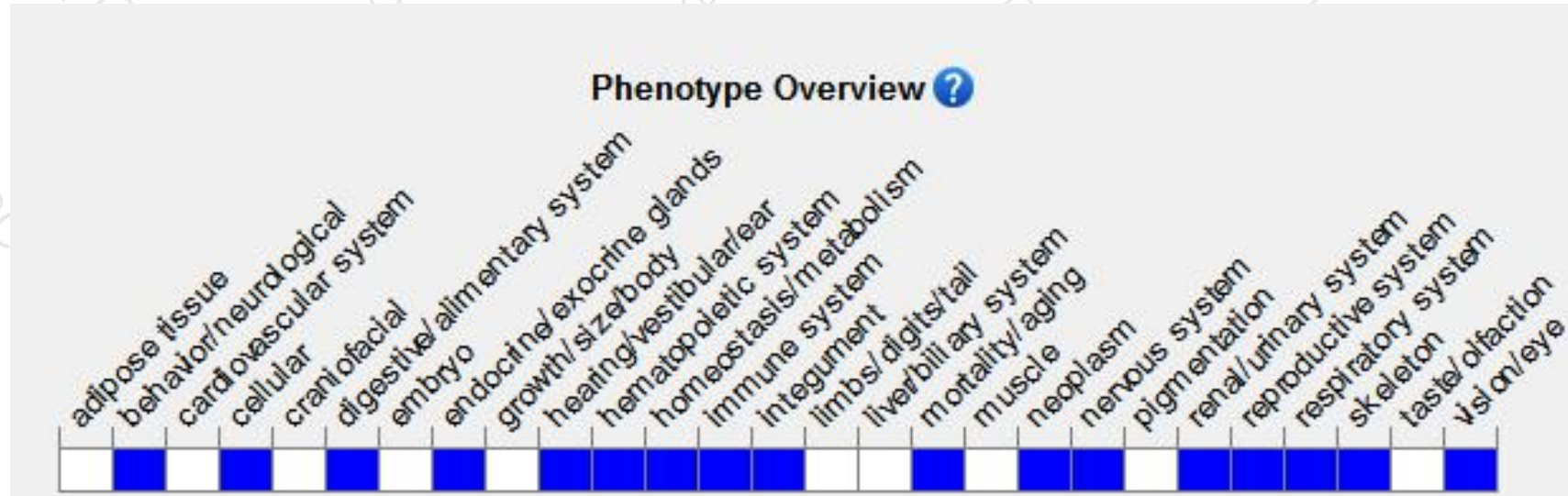◄ Reverse strand ———————————————— 90.96 kb ————————————————
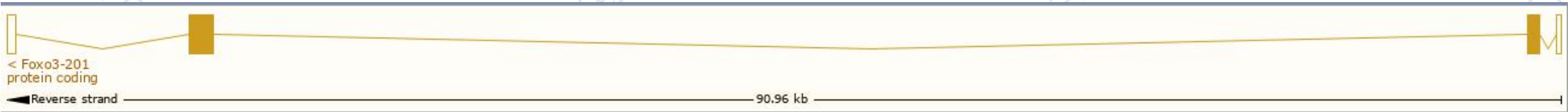
Genomic location distribution
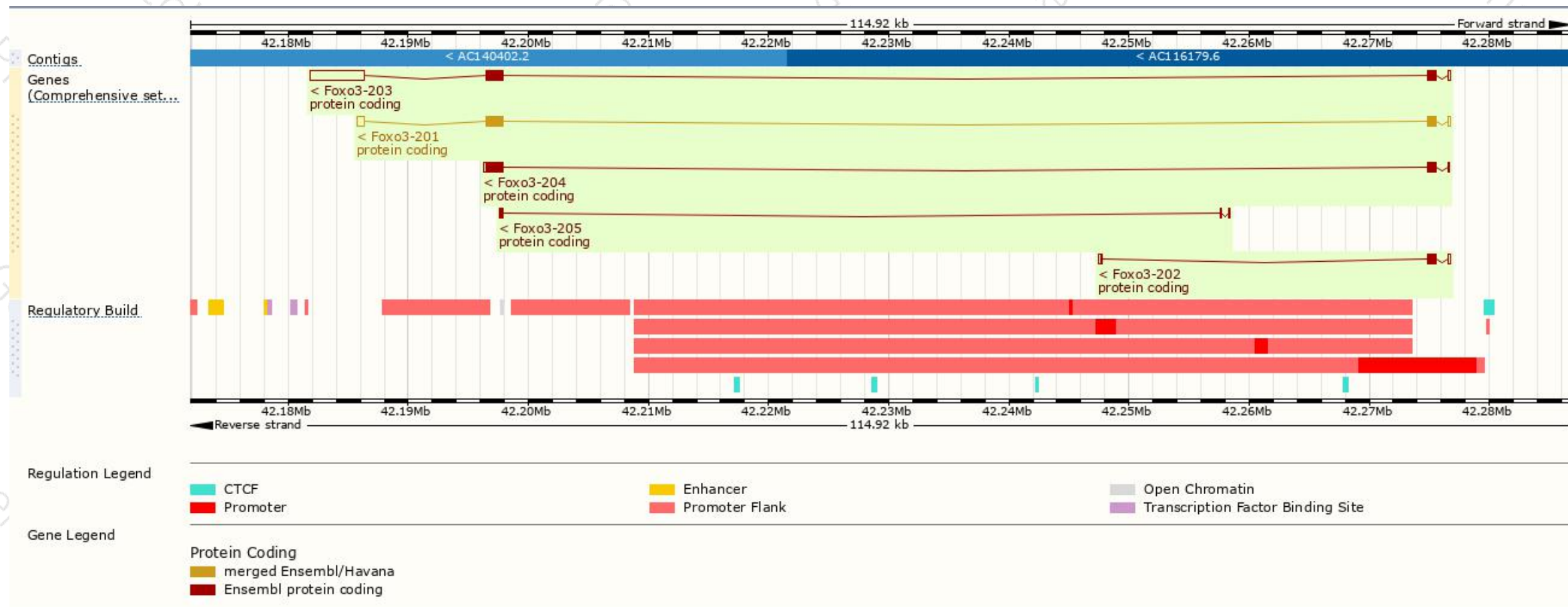
Protein domain

Mouse phenotype description(MGI)

Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/) .

Inactivation of the locus results in an ovarian defect involving follicular growth activation and leads progressively to female sterility. For some alleles defects in immune system function and hematopoiesis have also been reported.

If you have any questions, you are welcome to inquire.
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